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Poisoned Synapses not Crocodile Bile the Likely Killer of Mozambique Beer Drinkers

On January 11, 2015 news swept the globe reporting that scores of people died and 200 were sickened by drinking beer poisoned with crocodile bile in Mozambique. Thinking is now shifting to the possibility of poisoned synapses, not reptilian bile as the cause of these deaths.

A horribly tragic incident occurred at a funeral on January 9, 2015 in Mozambique, when people who had come to pay their last respects suffered poisoning from consuming the traditional home-made beer that was served. The mass fatalities of over 70 people devastated families, leaving many children orphans overnight. The toxic substance in the beer is still not known, but the media frenzy is astonishing. Is crocodile bile a deadly poison? How much bile can a person drink anyway? What is it in croc bile that could be so potent, trace amounts too weak to taste could kill and sicken hundreds of people?

Science writer for Forbes, David Kroll, traced the story back to the source. What he found was rumor and superstition, and what is more important, a likely cause of all this death. I refer you to his article for the details of his investigation and the suspicion that croc bile was not likely the poison in this brew.

The cause of these deaths is not yet known, but if it wasn't croc bile that killed the beer drinkers, let's have a look at the alternatives being suggested–all of them are related to nervous system function, and along the way delve a bit into the science of homebrew.

Not what made Milwaukee famous

The beverage brewed in Mozambique is a far cry from the Bud Light that most Americans associate with beer. Called *pombe*, the brew is a mixture of corn, bran, sorghum, and sugar, fermented with a different species of yeast (*Schizosaccharomyces pombe*) from the *Saccharomyces cerevisiae* used for making beer and wine. Beer as we know it is made from malted barley and hops. The fermented drink in Mozambique appears to somewhat resemble the corn-based traditional beverage, called *chicha*, which has been made in South America for centuries.

Professor Javier Carvajal Barriga, a microbiologist at Pontificia Universidad Católica del Ecuador in Quito, Ecuador, has resurrected yeasts from ancient fermentation vessels and other sources all over the world, including fermentation pottery from a pre-Incan tomb. From a fermentation vessel dating from 650 AD, Professor Carvajal Barriga resurrected dormant yeast and discovered an entirely new species, which he named *Candida theae*. This discovery also provided clues to how the ancient brew was made. Human spit was used to break down the corn starch into sugar that yeast can ferment into alcohol. (Saliva contains the starch digesting enzyme, amylase). His microbiological research also supported what conquistadores recorded about the traditional *chicha* recipe: that human feces were added to start the pot fermenting.

This is not how beer is made today. Malted barley is used, making spit unnecessary. Malting is a process whereby the grain is moistened and allowed to start sprouting. When a seed sprouts it generates enzymes to break down the starch to sugar, which is the energy store that will feed the sprouting plant. Beer makers hijack Nature's intentions by roasting the freshly sprouted barley to stop the germination process right after the enzyme is made. Then the brewer adds water to make a porridge that must be held at a precise sequence of temperature steps to coax the enzymes to make sugar from the starch. Yeast can then convert the sugar to alcohol and carbon dioxide. How early humans figured this all out is baffling, but no less amazing than the ingenuity used to make *chicha*.

So I when I heard of this mass death in Mozambique from poison *pombe*, I contacted Professor Carvajal Barriga for his insights.

"I'm really surprised by this news of poisonous beer," Carvajal Barriga says. In the middle ages they used rats, cats, a number of flowers and other elements like fungi to give magical properties to their brews. Some of those primitive beers would have been poisonous as well."

When I visited his lab in Quito in 2012, we shared a wonderful Belgian style ale that Carvajal Barriga had brewed from yeast he resurrected from an ancient oak vat that had held the first beer brewed in America. I remember him telling me how plants or mushrooms with psychoactive properties often would be added to the ancient *chicha*. (He could see this, by identifying species of yeasts in the ancient fermentation vessel that are associated with these other plants.) The ancient brew masters resorted to this because the species of yeast that they had to work with is killed by relatively low alcohol concentrations. So, unable to achieve the alcohol content of the beer we enjoy today, (outside the state of Utah), the bier meisters would toss in other substances to give the brew more of a kick.

"The poisoning in Mozambique may be due to chemicals added to the beer via flowers or even microtoxins produced by molds living on cereals," he suggests.

This is the same theory considered by Kroll in his Forbes article; specifically, the possibility that the foxglove plant might have been added to the Mozambique brew. Foxglove is the source of digitalis, a drug used for regulating heartbeat. The plant compound acts on the vagus nerve, part of the parasympathetic nervous system regulating heartbeat, and also acts directly on cardiac muscle, by changing the concentration of sodium ions inside these nervous system cells. The cellular voltage produced by neurons and muscle cells results from an imbalance in charged ions across the cell membrane–just as voltage is produced in a battery. Digitalis inhibits a membrane pump (sodium-potassium ATPase) that pushes sodium ions out of the cell and sucks potassium ions into the cell. When the pump is impaired by digitalis, sodium ions seep back in and the cellular voltage drops. This has a number of consequences, most importantly, changing the amount of calcium ions inside the cell. Calcium ions control many vital cellular processes, including the firing of synapses (i.e., release of neurotransmitter).

Poisoning synapses

Mass deaths at social gatherings often result from food poisoning, frequently caused by clostridial toxins made by bacteria found naturally in the soil, for example botulinum toxin. Botulinum toxin is one of the most deadly toxins known to man. The reason for this is that this toxin attacks the fundamental mechanism of communication in the brain–synaptic transmission.

When a nerve impulse reaches a nerve terminal, voltage-sensitive channels in the neuronal membrane open briefly and admit a spurt of calcium ions. These ions activate a protein called synaptotagmin, which is part of a complex machinery of molecules that tether synaptic vesicles next to the cell membrane of the nerve terminal. Synaptic vesicles are like submicroscopic “water balloons” filled with neurotransmitters, small molecules of various types that carry messages between neurons. When stimulated by an electrical impulse, this molecular machinery causes the synaptic vesicles to fuse to the cell membrane and rapidly dump its contents out of the neuron to stimulate receptors on the next neuron in the circuit.

Botulinum toxin (and tetanus toxin), cut the synaptic vesicle release proteins, thereby blocking synaptic transmission. (Either the synaptic proteins synaptobrevin, SNAP-25, or syntaxin can be attacked, depending on the specific type of Botulinum toxin.) The consequences of cutting communication lines in the nervous system are catastrophic, and food poisoning from clostricial toxins can kill quickly.

But beer is not often a source of food poisoning. This is because the process of brewing beer, unlike the process of making wine, requires prolonged boiling. Even a dead rat would become sterilized in the process of brewing beer. The poisoning in Mozambique most likely attacked synapses, but in a different manner.

Organophosphates

The extraordinary potency of clostridial toxins inspired the creation of man-made neurotoxins that work in a similar way for use in warfare against other people (nerve gas) or against insects (pesticides). At this point, this explanation seems the most likely cause of the mass death among the beer drinkers in Mozambique, according to the Forbes article. The brew was concocted in large barrels. If these or other utensils used in brewing, had been recycled from drums with pesticide residue in them, even low levels of organophosphates could have proven fatal.

Organophosphate pesticides work by inhibiting neurotransmission at synapses that use the nerurotransmitter acetylcholine to communicate. This includes the synapses called neuromuscular junctions that control our muscle contractions. Once acetylcholine is released from the nerve terminal and activates the muscle, the chemical signal to contract muscles must be terminated. This is accomplished by an enzyme in the synapse (acetylcholinesterase), which rapidly breaks down acetylcholine. Organophosphate pesticides and Sarin nerve gas prevent this enzyme from working. Unable to control muscles for breathing and other essential bodily functions that are controlled by synapses using acetylcholine, the victim (bug or enemy) dies quickly and quite miserably.

This brings the Mozambique tragedy close to home. An FDA analysis determined that 49% of fruit, 29% of vegetables, and 26% of grain products produced in the United States have pesticide residue. 50% of shallow water wells have pesticide contamination at detectable levels. Children are especially vulnerable to pesticide exposure because their smaller bodies make the same amount of pesticide on one apple a much higher dose than for an adult. Also, children crawl on the floor, play in dirt and put things in their mouths, increasing their exposure to pesticides. Organophosphate pesticide levels in people living in agricultural areas are higher than in people living in urban areas, but in a large sample of children between ages 8-15 in the general population, the level of pesticide residue in urine samples correlates directly with a risk for attention deficit hyperactivity disorder (ADHD).

In a 2012 review article in the journal *Pediatrics* the authors conclude that organophosphate exposures that are being experience by US children in the general population may have adverse neurodeveopmental consequences. The consequences of organophosphate exposure to the fetus and young children are known to include decreased IQ, ADHD, neurodevelopmental disorders, and cancer. For example, a study of Latino farm worker families from agricultural regions showed that organophosphate levels in urine from pregnant women were associated with lower IQ in their children at 7 years of age. Finally, many veterans of the Gulf War suffered chronic illness, which appears to be related to low-level exposure to Sarin gas and organophosphate pesticides they were exposed to on the battlefield.

We can only hope that the cause of the poisoning in Mozambique can be found, and that we learn from the tragedy.

Figure Caption An ale made by infusion mash of English pale malted barley, crystal and dextrin malt, and Monteuka, Nelson Sauvin and Green Bullet hops from New Zealand and Nottingham Ale yeast.

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R. Douglas Fields is Chief of the Nervous System Development and Plasticity Section at the National Institutes of Health, NICHD, in Bethesda, Maryland, and author of the popular book about glia The Other Brain published by Simon and Schuster. Dr. Fields is a developmental neurobiologist with a long-standing interest in brain development and plasticity, neuron-glia interactions, and the cellular mechanism of memory. He received degrees from UC Berkeley, San Jose State University, and UC San Diego. After postdoctoral fellowships at Stanford and Yale Universities he joined the NIH in 1987. Dr. Fields also enjoys writing about neuroscience for the general public. In addition to serving on editorial boards of several neuroscience journals, he serves as scientific advisor for Odyssey and Scientific American Mind magazines. He has written for Outside Magazine, The Washington Post Magazine, Scientific American and Scientific American Mind, and he publishes regularly for The Huffington Post, Psychology Today, and Scientific American on-line.

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